

Amendments to the Claims:

This listing of claims will replace all prior versions, and listings, of claims in this Application:

Listing of Claims:

1. – 103. (Canceled).

104. (Currently amended) An enucleation MEMS device for enucleating a cell comprising:

- (a) a substrate ~~for supporting the element of the device;~~
- ~~(b) —~~ comprising at least one well for holding a cell wherein the well comprises;
- (i) an enucleation penetration member to penetrate the cell to access the nucleus; and
- (ii) an enucleation pit for isolating the nucleus from the cell.

105. (Currently amended) The enucleation/nuclear transfer MEMS device of claim 104 wherein the well comprising at least one hollow protuberance is an emitter.

106. (Original) The enucleation/nuclear transfer MEMS device of claim 105 wherein the enucleation penetration member is an emitter.

107. (Currently amended) An enucleation/nuclear transfer MEMS device kit comprising:

- (a) a ~~base~~ second substrate comprising an input well for depositing a cell, a lever element for controlling the cell apposition and a micropump for handling fluids; and
- (b) an enucleation/nuclear transfer MEMS device of claim 105.

108. (Previously presented) An enucleation MEMS device kit for enucleating a cell comprising:

- (a) a centrifugal platter for supporting at least one MEMS device and for applying a centripetal force to a cell or group of cells contained within the MEMS device wherein the centrifugal disk comprises a plurality of ports for affixing the MEMS devices and a securing means for securing the centrifugal disk to a spinner or driving means; and
- (b) at least one enucleation MEMS device of claim 104.

109. (Currently amended) The enucleation MEMS device kit of claim ~~107~~ 108 wherein

(a) the centrifugal platter comprises a plurality of grooves arranged in a concentric pattern and wherein each groove has an inner and outer edge;

(b) at least one enucleation MEMS device is bonded to the outer edge of a groove in an orientation such that the axis of each well of the enucleation MEMS device is horizontal to the plane of the centrifugal platter; and

(c) the inner edge of the grooves forming divided compartments comprising a single well which restrict the movement of materials from one compartment containing a single well to another compartment.

110. (Currently amended) A method of using the enucleation MEMS device kit of claim 108 comprising the steps of:

(a) filling the grooves of the centrifugal platter with a fluid;

(b) loading the fluid within the grooves of the centrifugal platter with at least one oocyte or embryo;

(c) rotating the kit such that centripetal forces are applied to the centrifugal platter such that the oocyte or embryo are thrust against the wall of the well such that the enucleation penetration member of the enucleation MEMS device penetrates the surface of the oocyte or embryo and

(d) extruding the cell contents out of the oocyte or embryo containing a nucleus into the enucleation pit forming a cell extrusion; and

(e) severing any remnant of the cell extrusion in enucleation pit with a slideable shutter.

111. (Currently amended) A method of using the enucleation MEMS device kit of claim 108 comprising the steps of:

(a) filling ~~the~~ a plurality of grooves of the centrifugal platter with a fluid;

(b) loading the fluid in the grooves of the centrifugal platter with at least one oocyte or embryo;

(c) applying centripetal forces to the kit such that the oocyte or embryo makes contact with the enucleation MEMS device and penetrates the surface of the oocyte or embryo;

(d) extruding a portion of the oocyte or embryo contents into the enucleation pit to isolate the nucleus; and

(e) severing any remnant of cell extruded into enucleation pit using a slideable shutter.

112. (Currently amended) An enucleation MEMS device for enucleating a cell comprising:

(a) a base substrate comprising;

(i) an input well to introduce a cell;

(ii) a lever to control the motion of the cell; and

(iii) a pump for applying a force to extrude a portion of the cell; and

(b) an enucleation MEMS device.

113. (Canceled).

114. (Currently amended) ~~(a)~~—A method of making an enucleation/nuclear transfer MEMS device comprising the steps of:

(a) depositing a first mask on the top surface of a substrate inscribing a square shape;

(b) etching the first mask to form a plurality of wells;

(c) depositing a second mask in the wells of step (b) such that an enucleation penetration member is inscribed at the bottom of each well;

(d) etching the second mask ~~(g)~~ to form the enucleation penetration member;

(e) applying a third mask within each well adjacent to the enucleation penetration member such that an enucleation pit is inscribed;

(f) etching the third mask ~~(i)~~ to form the enucleation pit;

(g) applying a fourth mask such that a slidable shutter is inscribed;

(h) etching the fourth mask ~~(k)~~ to form the slideable shutter; and

(i) depositing a circuit lead to provide communication between the shutter and a controller.

115. (Currently amended) An enucleation/nuclear transfer MEMS array for the enucleation and transfer of a donor nucleus or donor cell comprising ~~comprising~~:

- (a) a central loading manifold for the loading of a donor nucleus or donor cell into the device;
- (b) at least one well for holding an oocyte or embryo during the enucleation process, wherein the well comprises:
 - (i) a hollow protuberance in the well for penetrating the oocyte or embryo to introduce a donor nucleus or donor cell;
 - (ii) an enucleation penetration member for penetrating an oocyte or embryo to facilitate the removal of a cell nucleus;
 - (iii) an enucleation evacuation siphon to provide suction to remove the nucleus from the oocyte or embryo forming an enucleated oocyte or embryo; and
- (c) a dynamic hydropressure column for providing a pressurized fluid to introduce the donor nucleus or donor cell through the hollow protuberance into the enucleated oocyte or embryo.

116. (Currently amended) A method of using enucleation/nuclear transfer MEMS array of claim 14 comprising the steps of:

- (a) loading a donor nuclei or donor cells into central loading manifold;
- (b) filling the input well with a fluid;
- (c) loading the input well with an oocyte or an embryo; and
- (d) rotating the enucleation/nuclear transfer MEMS device.

117. (Currently amended) ~~rotating the enucleation/nuclear transfer MEMS device.~~ A method of making an enucleation/nuclear transfer MEMS device comprising the steps of:

- (a) etching a plurality of parallel channels on a first side of a plurality of silicon wafers in which the wafers each have a second unetched side;
- (b) silicon fusion bonding the unetched side of a plurality of silicon wafers of step (a) to the etched side of a plurality of silicon wafers of step (a) such that the etched channels are in parallel to form a mega-laminate wherein the mega-laminate has a plurality of channels;
- (c) cutting the mega-laminate at an angle perpendicular to the long axis of the etched channels thereby forming a slice of the mega-laminate having a top surface and a bottom surface wherein each surface exposes an end of the channel;

- (d) silicon fusion bonding the bottom surface of the slice of the mega-laminate to the etched side of a channel-etched base-plate wafer;
- (e) depositing a first mask on the top surface of the slice of the mega-laminate such that a region surrounding each channel end is free of mask;
- (f) etching the mask to form a plurality of wells;
- (g) depositing a second mask in the wells of step (f) such that an enucleation penetration member is inscribed at the bottom of each well;
- (h) etching the second mask ~~(g)~~ to form the enucleation penetration member;
- (i) applying a third mask within each well adjacent to the enucleation penetration member such that an enucleation pit is inscribed;
- (j) etching the third mask ~~(i)~~ to form the enucleation pit;
- (k) applying a fourth mask such that a slidable shutter is inscribed;
- (l) etching the fourth mask ~~(k)~~ to form a slideable shutter; and
- (m) depositing a circuit lead to provide communication between the slideable shutter and the controller.

118. (Currently amended) The method of making an enucleation/nuclear transfer MEMS device of claim ~~116~~ further ~~117~~ further comprising applying a coating to the mega-laminate top surface after step (h).

119. (Original) The method of making an enucleation/nuclear transfer MEMS device of claim 117 wherein the coating is a polypeptide, peptide or protein.

120. (Currently amended) The method of making an enucleation/nuclear transfer MEMS device of claim ~~118~~ 119 wherein the polypeptide is polylysine.

121. (Currently amended) An enucleation/nuclear transfer MEMS device kit for enucleating a cell comprising:

- (a) a centrifugal platter for providing support for at least one MEMS device and for applying a centripetal force to a cell or group of cells contained within the MEMS device wherein the centrifugal disk comprises a plurality of ports for affixing the MEMS ~~devices~~ device and a securing means for securing the centrifugal disk to a spinner or driving means; and

(b) at least one enucleation/nuclear transfer MEMS device ~~comprising of claim 1~~ a substrate comprising at least one well for holding a cell wherein the well comprises:

- (i) an enucleation penetration member to penetrate the cell to access the nucleus; and
- (ii) an enucleation pit for isolating the nucleus from the cell.

122. (Currently amended) The enucleation/nuclear transfer MEMS device kit of claim ~~120~~ 121 wherein the enucleation/nuclear transfer MEMS device is permanently affixed to the centrifugal platter.

123. (Previously presented) The enucleation/nuclear transfer MEMS device kit of claim 121 wherein

(a) the centrifugal platter comprises a plurality of grooves arranged in a concentric pattern and wherein each groove has an inner and outer edge;

(b) at least one enucleation/nuclear transfer MEMS device is bonded to the outer edge of a groove in an orientation such that the axis of each well of the enucleation/nuclear transfer MEMS device is horizontal to the plane of the centrifugal platter; and

(c) the inner edge of the grooves forming divided compartments comprising a single well which restrict the movement of materials from one compartment containing a containing a single well to another compartment.

124. (Currently amended) A method of using the enucleation/nuclear transfer MEMS device kit of claim ~~122~~ 121 comprising the steps of:

(a) filling ~~the~~ a plurality of input manifold wells of at least one enucleation/nuclear transfer MEMS device with a fluid;

(b) loading the fluid-filled wells of step (b) with at least one oocyte or embryo; and

(c) rotating the kit and thus applying a centripetal force on the enucleation/nuclear transfer MEMS/centrifugal platter.

125. (Currently amended) A method of using an enucleation/nuclear transfer MEMS device kit of claim ~~122~~ 123 comprising the steps of:

(a) loading donor nuclei or cells into the central loading manifold;

(b) filling the grooves of the centrifugal platter with a fluid;

(c) loading the grooves of the centrifugal platter with at least one oocyte or embryo;

(d) applying a centripetal force to the kit whereby the oocyte or embryo makes contact with the pertubance of the enucleation/nuclear transfer MEMS device and the pertubance penetrates the surface of the oocyte or embryo; and

(e) removal of oocyte or embryo from the kit.

126. (Original) A centrifugal platter for supporting MEMS devices and for applying a centripetal force to a cell or group of cells contained within a MEMS device facilitating migration of the cells onto the enucleation region of the MEMS device wherein the centrifugal platter comprises a circular disc, a plurality of ports for holding the MEMS devices, and a securing means for attaching to a spinner or driving means.

127. (Original) A Cytoplasmic Transfer MEMS array for the transfer of cytoplasm from one cell to another comprising:

(a) a cytoplasmic transfer MEMS device comprising a substrate comprising:

(i) at least one first well wherein each first well compromises a hollow protuberance;

(ii) at least one second well wherein each second well compromises a hollow protuberance;

(iii) an extraction siphon in fluid communication with the hollow protuberance in the first well and with the hollow protuberance in the second well;

(iv) a supplemental input channel in fluid communication with the extraction siphon; and

(b) a centrifugal platter having a top and a bottom surface wherein the cytoplasmic transfer MEMS device is attached to the top surface.

128. (Original) A Cytoplasmic Transfer MEMS array further comprises:

(a) a cytoplasmic transfer MEMS device comprising a substrate comprising:

(i) at least one first well wherein each first well compromises a hollow protuberance;

(ii) at least one second well wherein each second well compromises a hollow protuberance;

(iii) an extraction siphon in fluid communication with the hollow protuberance in the first well and with the hollow protuberance in the second well;

(iv) a supplemental input channel in fluid communication with the extraction siphon; and (b) a centrifugal platter for applying a centripetal force to a cell or group of cells contained within a MEMS device and comprises a circular disk having a plurality of ports for holding the MEMS device.

129. (Currently amended) A method of using a Cytoplasmic Transfer MEMS ~~device~~ array of claim 128 comprising:

- (a) loading fluid in the extraction siphon;
- (b) loading a cytoplasmic donor oocyte or embryo in the first well;
- (c) loading a recipient oocyte or embryo in the second well; and
- (d) applying a force to the Cytoplasmic Transfer MEMS device by rotating the

Cytoplasmic Transfer MEMS array on a spinner or driving means.

130. (Currently amended) A multi-layer cell culture MEMS array for culturing a cell or groups of cells, comprising a multi-laminate planar layer comprising:

(a) at least one loading compartment for loading cells or groups of cells or fluids into the ~~device~~ array;

(b) at least one enclosed channel in fluid communication with the loading compartment and wherein the enclosed channel allows for the passage of cells;

(c) at least one movement track attractive to labelable zona anchor MEMS attached to the enclosed channel;

(d) at least one removal compartment for the removal of cells or groups of cells; and

(e) at least one circuit lead providing communication between at least one movement track and a controller unit.

131. (Previously presented) A multi-layer cell culture MEMS array of claim 130 further comprising at least one router element which resides on a movement track.

132. (Currently amended) The multi-layer cell culture MEMS array of claim 131 wherein at least one enclosed channel with movement track is in fluid communication with a

culture manifold for the transport of a cell or group of cells and fluid through the ~~cell culture device~~ multi-layer cell culture MEMS array.

133. (Original) A single-layer MEMS culture array for culturing cells or a group of cells comprising:

(a) at least one loading compartment for loading cells or groups of cells or fluids into the device;

(b) at least one enclosed channel in fluid communication with the loading compartment and wherein the enclosed channel allows for the passage of cells;

(c) at least one movement track attractive to labelable zona anchor MEMS attached to the enclosed channel;

(d) at least one removal compartment for the removal of cells or groups of cells; and

(e) at least one circuit lead providing communication between at least one movement track and a controller unit.

134. (Currently amended) A single layer cell culture MEMS array of claim ~~135~~ 133 further comprising at least one router element which resides on a movement track.

135. (Currently amended) The single layer cell culture MEMS array of claim ~~136~~ 133 wherein at least one enclosed channel with movement track is in fluid communication with a culture manifold for the transport a cell or group of cells and fluid through the cell culture device.

136. (Previously presented) The enucleation MEMS device of claim 104 further comprising:

(a) a slidable shutter adjacent to the union between the enucleation penetration member and the enucleation pit for severing a portion of the cell containing the nucleus; and

(b) a controller in communication with the slideable shutter through a circuit lead to control the movement of the slideable shutter.

137. (Previously presented) A method of using the enucleation MEMS device of claim 105 comprising the steps of:

(a) filling the well with a fluid;

(b) loading the well with an oocyte or embryo;

(c) applying a centripetal force to the enucleation MEMS device thereby causing the cell to be forced against the enucleation penetration member and forcing a portion of the cell containing the nucleus into the enucleation pit.

138. (Previously presented) The enucleation MEMS device kit of claim 107 wherein the enucleation MEMS devices are permanently attached to the centripetal platter.